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PHYTOCHEMICAL ANALYSIS OF PLANT SPECIES OF GENUS ZANTHOXYLUM

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ABSTRACT

The present article illustrates phytochemical investigations carried out on three plant species i.e. Zanthoxylum armatum (D.C), Zanthoxylum oxyphyllum (Edgew) and Zanthoxylum rhetsa (Roxb.) (D.C). Percent yield of the crude ethanolic extracts from Zanthoxylum armatum (Leaves), Zanthoxylum oxyphyllum (Leaves), Zanthoxylum oxyphyllum (Seeds), Zanthoxylum rhetsa (Leaves), Zanthoxylum rhetsa (Seeds) were 7.78%, 9.85%, 14.1%, 9.23%, and 14.21% respectively. The qualitative analysis showed the abundance of Glycosides, Coumarins, Flavonoids, Phenols, and Tannins in crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL, and ZRS. Thus, the preliminary phytochemical screening revealed the presence of medicinally important constituents in the plants studied. Phenolic content was found maximum in leaves of Zanthoxylum oxyphyllum followed by leaves of Zanthoxylum armatum. Seeds of Zanthoxylum rhetsa showed maximum concentration of Flavonoid followed by leaves of Zanthoxylum armatum. The present qualitative and quantitative analysis explores the wider possibility of efficient and productive extraction of different secondary metabolites from crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL, and ZRS.

KEYWORDS: Phytochemical Screening, Zanthoxylum armatum (D.C), Zanthoxylum oxyphyllum (Edgew) and Zanthoxylum rhetsa (Roxb.) (D.C)

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Original Article

INTRODUCTION

The potential of plants to serve human beings in a range of aspects has been well documented since antiquity (Albuquerque *et al.*, 2011; Qureshi *et al.*, 2014; Sangeetha and Baskar, 2015). Plant products have been part of Phytomedicines since time immemorial (Criagg and David, 2001). A/c to Wadood *et al.*, (2013), presence of phytochemicals is considered as active medicinal chemical constituents. Medicinal plants play a vital role in preventing various diseases (Wadood *et al.*, 2013) and their medicinal value lies in some bioactive constituents (like flavonoids and phenols) that produce a definite physiological action on the human body (Mir *et al.*, 2013). Phenols have antibacterial, antifungal, nematicidal, insecticidal, antioxidant, antimutagenic, and other activities (Oksana *et al.*, 2012).

A/c to Brijwal *et al.*, (2013), India has richest plant based medicinal traditional system because of its rich biodiversity. Exploration of medicinal plants in the North east India is a matter of attraction to the scientific fertinity, traders as well as pharmaceuticals (Shankar and Rawat, 2013). Chakraborty *et al.*, (2012) emphasized on the need of detailed and systematic review on ethanobotanical studies in the North East India. Many species of *Zanthoxylum* are used in traditional medicine especially in America, Africa, and Asia (Prieto *et al.*, 2010). Species of *Zanthoxylum* Genus possess high medicinal, economical as well as ecological importance (Brijwal *et al.*, 2013)

coupled with significant insecticidal and antifungal activities and it is a promising source of various secondary metabolites (Patino *et al.* 2012). Therefore, efforts were made to characterize the different types of metabolites presented in selected species of *Zanthoxylum* Genus followed by their quantitative estimation based on preliminary phytochemical screening.

RESEARCH METHODOLOGY

Leaves of *Zanthoxylum armatum* (ZAL), *Zanthoxylum oxyphyllum* (ZOL), *Zanthoxylum rhetsa* (ZRL) and Seeds of *Zanthoxylum oxyphyllum* (ZOS), *Zanthoxylum rhetsa* (ZRS) were collected from Chungliyimsen village, District-Mokokchung, Nagaland (North-East India). Plant materials were cleansed with distilled water and allowed to dry for 15 days in dark. Thereafter, it was coarsely crushed using homogenizer and grinded mechanically of mesh size 1 mm. The powdered plant material was extracted successively with 70% Ethanol. After 48 hours, extracts were filtered by using muslin (cheese cloth) followed by Whatman filter paper No.1 and filtrates were evaporated to dryness and weighed. The crude extracts were stored in air tight glass containers at 4°C till further analysis. All extracts were subsequently stored at -20 ^{0}C in deep freezer until bioassays were conducted. Solvents used in extraction and Reagents for phytochemical analysis was of pure analytical grade.

Phytochemical investigations of crude alcoholic extracts of ZAL, ZOL, ZRL, ZOS, and ZRS include: Percent of yield of Extract (Patil and Gaikwad, 2010); Preliminary Phytochemical screening (Harbone, 1998; Fransworth, 1996; Rangari, 2002; Khandelwal, 2005; Sofowara, 1993; Trease and Evans, 1989; Harborne, (1973); Total Phenolic Content (Singleton *et al.*, 1999) and Total Flavonoid Content (Nguyen and Eun, 2011).

RESULTS AND DISCUSSIONS

Percentage Yield of Extract

Percent yield of the crude ethanolic extracts from *Zanthoxylum armatum* (Leaves), *Zanthoxylum oxyphyllum* (Leaves), *Zanthoxylum oxyphyllum* (Seeds), *Zanthoxylum rhetsa* (Leaves), *Zanthoxylum rhetsa* (Seeds) were 7.78%, 9.85%, 14.1%, 9.23%, and 14.21% respectively. Highest yield was observed in seeds of *Zanthoxylum rhetsa* followed by seeds of *Zanthoxylum oxyphyllum.* Similar results were also reported in the studies conducted on different species of *Zanthoxylum* using different plant parts. 1.45% of extract yield was observed by Islam *et al.*, (2014) in 95% crude ethanolic extract of *Zanthoxylum budrunga* seeds. Reddy and Jose (2011) reported 1.83% of extract yield from aqueous extract of *Zanthoxylum rhetsa* seeds, *while* Buragohain *et al.*, (2011) reported 1.22 % of extract yield in methanolic extract of *Zanthoxylum oxyphyllum* tender shoots. Present findings were well correlated with the findings of Verma and Khosa (2012).

Preliminary Phytochemical Analysis

Phytochemical screening is a preliminary step in the characterization of plant products. Preliminary screening for presence or absence of a particular class of compound is qualitative in nature and form a robust base for the quantitative estimation of bioactive components. In the present study, phytochemical components in crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL, and ZRS were investigated through a series of chemical tests. Phytochemicals were screened for Alkaloids, Carbohydrates, Glycosides, Coumarins, Proteins & Amino acids, Phytosterols, Phenolic compounds, Tannins, Flavonoids, Anthocyanins, Lignins, Terpenoids, Phlobatannins and Quinones.

The preliminary phytochemical screening had showed the abundance of Glycosides, Coumarins, Flavonoids, Phenols, and Tannins in crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL, and ZRS. However, Phytosterols, Anthocyanins and Phlobatannins were not detected in all the tested samples. It was observed that the ZOS and ZRS showed abundance of Flavonoids and Coumarins followed by Phenols and Tannins. ZAL, ZOL and ZRL showed significant presence of Tannins followed by Glycosides. Crude ethanolic extract of leaves of *Zanthoxylum armatum* showed abundance of Flavonoids, Glycosides, Tannins and traces of Phenols, Carbohydrates, Coumarins and Lignins. Ethanolic extract of ZOL was screened positive for Phenols, Tannins, Coumarins and Glycosides. Considerable presence of Phenols, Flavonoids and Coumarins followed by Alkaloids, Carbohydrates, and Tannins with traces of Glycosides and Quinones were screened in the crude ethanolic extract of ZOS. Maximum number of secondary metabolites was screened in crude ethanolic extract of ZRS with abundance of Flavonoids, Glycosides, Coumarins, Proteins & amino acids, Tannins followed by Phenols, Alkaloids, Terpenoids, with traces of Carbohydrates and Quinones. Crude ethanolic extract of ZRL was found positive in preliminary screening for Glycosides, Phenols, Flavonoids, Coumarins and Tannins. The present qualitative analysis explores the wider possibilities for efficient and productive extraction of different secondary metabolites from crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL, and ZRS.

Estimation of Secondary Metabolites

Plant phenolics are the major group of chemical species that act as primary antioxidants (Hatano *et al.*, 1989) and have high redox potentials which allow them to act as oxygen quenchers (Kahkonen *et al.*, 1999) and are responsible for multiple biological effects (Pise *et al.*, 2010).

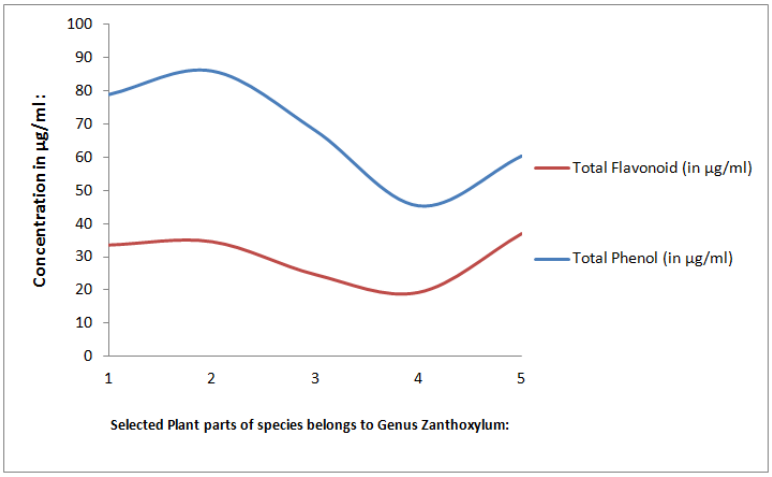

Figure 1: Comparative Analysis of Total Phenol and Flavonoid Content in Selected Plant Parts of *Zanthoxylum* Species Using Ethanol as a Solvent

- *Zanthoxylum armatum* (Leaves); 2) *Zanthoxylum oxyphyllum* (Leaves); 3) *Zanthoxylum oxyphyllum* (Seeds); 4) *Zanthoxylum rhetsa* (Leaves); 5) *Zanthoxylum rhetsa* (Seeds).

Figure 1 shows variation in Total Phenol and Flavonoid content. Phenolic content was found maximum in ethanolic extract of *Zanthoxylum oxyphyllum* leaves followed by *Zanthoxylum armatum* leaves. However, minimum concentration of Phenol was observed in ethanolic extract of *Zanthoxylum rhetsa* leaves. Ethanolic extract of *Zanthoxylum rhetsa* seeds showed maximum concentration of Flavonoids followed by the leaves of *Zanthoxylum armatum*. While analysing the trend in Phenol and Flavonoid content in the leaves and seeds of *Zanthoxylum oxyphyllum*, it was observed

that the ethanolic extract of their leaves were showed higher concentration in comparison to seeds. However, in case of *Zanthoxylum rhetsa*, the trend was opposite. In this case, it was observed that the content of Phenol and Flavonoid was higher in ethanolic extract of seeds instead of leaves. Ethanolic extract of *Zanthoxylum oxyphyllum* leaves analysed with higher Phenolic and Flavonoid content in comparison to their seeds. Previous studies also reported significant yield in Total Phenol and Flavonoid contents from crude extracts of Zanthoxylum species. Prabhash *et al.,* (2014) found 62.3 µg/ml of Total Phenol from methanolic extracts of *Zanthoxylum rhetsa* Leaves. A/c to Mahadkar *et al.,* (2013), *Zanthoxylum rhetsa* fruits with methanolic extracts yields 0.061±0.29 g/100g FW of Phenols. *Zanthoxylum rhetsa* bark with ethyl acetate and butanol fractions yields 20.47±0.09 and 14.14±0.185 mg GAE/g DW of Phenols, respectively (Santhanam *et al.,* 2013). Vidyamadhavi *et al.,* (2014) found the Flavonoid yield of 258 µg, 533 µg and 349 µg equivalent of tannic acid (Vidyamadhavi *et al.,* 2014) in ethyl acetate, methanol and aqueous extracts of aerial parts of *Zanthoxylum rhetsa. Zanthoxylum armatum* leaves extract yields 28.46 mg/L gallic acid equivalent of Phenols (Mahadkar *et al.,* 2013). Statistical analysis was also employed in order to estimate the correlation between Phenol and Flavonoid content among the selected plant parts (Leaves or Seeds) of *Zanthoxylum armatum, Zanthoxylum oxyphyllum*, and *Zanthoxylum rhetsa* using ethanol as a solvent. The strongest correlation was observed in total phenolic and total flavonoid content of *Zanthoxylum oxyphyllum* and *Zanthoxylum rhetsa* leaves and seeds.

CONCLUSIONS

Traditional medicine is pervasively used by nearly 80% of the population in African countries (WHO, 2000). In China, 40% of health care depends on traditional medicine (WHO, 1999). In India there are about 45,000 plant species, out of them several thousands have been claimed to possess medicinal properties (Grover *et al.,* 2002). A/c to the MOEF (GOI) report, the herbal industry in India uses about 8000 medicinal plants (Gundimeda *et al.*, 2006; Kumar and Janagam, 2011). Another literature estimates that in India there are 880 medicinal plants species involved in all India trade out of which about 48 species are exported (EXIM; Gundimeda *et al.*, 2006; Kumar and Janagam, 2011). The international market turnover for herbal products is of $6.2 billion; however India's share in the global medicinal plants related export trade is just 0.5 percent (WHO, 2002; Kumar and Janagam, 2011). In order to materialize the biodiversity rich potential, India must focus and develop the scientific cultivation, post harvest technology, processing, manufacturing, patenting, marketing, etc., for medicinal plants, as suggested by Singh, (2006).

A/c to Wadood *et al.,* (2013), the phytochemical analysis of the medicinally important plants has commercial interest in both research institutions and pharmaceutical organizations (Wadood *et al.,* 2013). The present phytochemical study investigates qualitative screening and quantitative analysis of *Zanthoxylum armatum* (D.C), *Zanthoxylum oxyphyllum* (Edgew) and *Zanthoxylum rhetsa* (Roxb.) (D.C) plants. Preliminary Phytochemical screening showed the presence of Alkaloids, Carbohydrates, Glycosides, Coumarins, Proteins & Amino acids, Phenolic compounds, Tannins, Flavonoids, Lignins, Terpenoids and Quinones in crude ethanolic extracts of ZAL, ZOL, ZOS, ZRL and ZRS. The results of phytochemical analysis revealed the presence of medicinally important constituents in the plant species studied. The present study strongly recommended the further work to isolate, purify, characterize, and standardize the bioactive constituents responsible for the medicinal properties of plants belongs to Genus *Zanthoxylum.*

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